Fruit Characteristics and a Simple Estimate of Oil Content in Twenty Six Olive Genotypes in Argentina

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ABSTRACT

There are more than two thousand varieties of olives grown worldwide, most of them native to the Mediterranean Region. However, in Argentina, only a small number of olive varieties are cultivated. In Mendoza Province, there is a collection of olive varieties that includes more than seventy accessions; nevertheless, little is known about the adaptation of the different varieties to the province's arid conditions. The aims of this work were to evaluate fruit characteristics of agronomic importance in 26 olive accessions of the germplasm collection and to compare the fruit characteristics of local variety 'Arauco' in different environments and growing seasons in Mendoza. In addition, an economic, quick, and easy method to estimate fruit oil content was calibrated and validated using a wide range of olive varieties. Varieties and years showed significant ($P \leq 0.01$) differences for all the evaluated characteristics. Fruit oil content was closely and positively related to pulp dry weight ($y= -0.05+0.56x$; $r= 0.99$). The varieties highlighted from the collection for their high fruit oil concentration in fresh base and low moisture were 'Canino', 'Cornezuelo', 'Cucci', 'Dritta', 'Dulzal', 'Farga', 'Frantoio', 'Grappollo', 'Nebbio', 'Picual', and 'Villalonga'. Additionally, all fruit characteristics evaluated in 'Arauco' were similar among the studied environments and were significantly ($P \leq 0.01$) influenced by seasonal conditions. The proposed model, calibrated and validated with independent data, would allow determining the best harvest time simply from pulp dry weight as model input, determining oil content and analyzing several samples in a short period of time.

Keywords: Arauco variety, Harvest time, Maturity index, Modelling fruit oil, Pulp/Stone ratio.

INTRODUCTION

There are more than two thousand varieties of olives grown worldwide, most of them native to the Mediterranean Region (Muzzalupo, 2012). In the last decades, the olive-planted area has increased greatly in the Mediterranean Basin and elsewhere, such as the American Continent. In Argentina, only a small number of olive varieties are cultivated (Vossen, 2007), and the characterization of more varieties is required because of two main reasons. Firstly, at the beginning of the 20th century,

European immigrants (mainly from Spain and Italy) brought a few olive varieties with good adaptation to Mediterranean conditions, but widely different from those of northwestern Argentina (Gómez Del Campo *et al*., 2010). These varieties have shown limited adaptation to various Argentine regions leading to low yield or bad quality oils (Vossen, 2007). For example, the 'Frantoio' variety showed low production in Catamarca Province (Aybar *et al*., 2015). In addition, 'Frantoio', 'Arbequina', 'Manzanilla', and 'Coratina' varieties showed low oleic acid content and high palmitic and linoleic acid contents in

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the northwestern provinces (Ceci and Carelli, 2007).

Secondly, Argentina's olive-growing areas have increased from 30,000 to 90,000 ha in the last three decades. New olive orchards are characterized by large areas, high plant density, and hedgerow training systems that encourage the use of few varieties (Tous *et al.*, 2010). Before the olive crop continues its expansion, it is crucial to determine which varieties adapt better to Argentinian conditions.

A large number of olive varieties has been evaluated in different environments of the Mediterranean Basin in terms of fruit characteristics, production, and oil quality (Muzzalupo, 2012; Rallo *et al.*, 2008). However, in Argentina, agronomic characterization has been limited to a small number of varieties with more commercial development (Torres *et al.*, 2017). The "Olive Germplasm Collection of Mendoza" is the most important collection of Latin America with more than 70 years of age and 74 accessions. Trentacoste and Puertas (2011) selected 25 olive varieties, scarcely spread in the olive groves of Mendoza, with low-moisture fruit and high oil concentration, i.e., fruits with a potential high oil yield in industrial terms. During olive oil extraction, fruits with high moisture and low oil concentration could be associated to the generation of "emulsions" (Aguilera *et al.*, 2010) reducing oil extraction.

The collection includes the autochthonous variety 'Arauco' and two of its clones, 'Criolla San Martín (SM)' and 'Criolla Salvarredi (S)' (Trentacoste and Puertas, 2011). In Mendoza, 'Arauco' is mainly destined to table olives that occupy 20% of the olive-cultivated surface (IDR, 2016). Little is known about the characteristics of 'Arauco' despite the importance of this variety for Argentine olive culture (Searles *et al.*, 2012). The study of autochthonous varieties in other regions and their comparison with non-native varieties has allowed valuing genetic resources (Lazović *et al.*, 2016).

In new olive orchards, there is a tendency to use mechanical harvest that has been accelerated through the use of new technologies (Zipori *et al.*, 2016) requiring faster decision-making. Thus, an evaluation of olive varieties requires a fast method to determine fruit maturity stage at maximum fruit oil concentration. Currently, many methods are used to determine oil concentration in olive fruits. Some of those methods require expensive equipment, toxic reagents, or both (e.g. Near-Infrared Reflectance Spectroscopy, Soxhlet, and Autelec). Avidan *et al.* (1999) developed a technique to determine fruit oil concentration without the use of expensive equipment. However, the technique is a laborious determination that requires at least four days to obtain results, dangerous reagents, and skilled labor.

In olive fruits, more than 90% of the oil is in the pulp (Beltrán *et al.*, 2003). According to Trentacoste *et al.* (2010), fruit oil concentration (%) [(g oil g^{-1} fruit dry weight)$\times$100] depends on fruit size, while fruit oil content (g oil fruit^{-1}) is the net oil content. Oil content could be indirectly estimated from Fruit Pulp Dry Weight (FPDW). However, FPDW and oil content are highly dependent on variety (Beltrán *et al.*, 2003) and environmental conditions (Arji, 2017; Mickelbart and James, 2003). Therefore, a more confident predictive model that explains the relationship between oil content and pulp dry weight should include a wide range of both genetic and environmental variability.

The aims of this work were to: (i) Evaluate fruit characteristics with agronomic significance in 26 olive accessions of the germplasm collection of Mendoza Province; (ii) Compare fruit characteristics of the local 'Arauco' variety in three environments during three growing seasons in Mendoza, and (iii) Calibrate and validate an economic, quick, and easy method to estimate fruit oil content using a wide range of olive varieties and environments.

MATERIALS AND METHODS

Sites and Plant Material

Experiment 1: Was carried out during 2014, 2015, 2016, and 2017 growing seasons evaluating a subset of twenty-six varieties from a collection planted at the experimental farm of INTA Junín (33∘ 06 S, 68∘ 29 W, 653 m asl) in the arid environment of Mendoza, Argentina. The area has an annual average temperature of 17.3ºC, an average rainfall of 275 mm (mainly in summer and far below crop water needs), and a frost-free period from November to April. The olive trees were seventy-years-old and were planted at a low density (12×12 m), under surface irrigation method. The 26 varieties evaluated included 23 varieties introduced from Spain and Italy and three 'Arauco' clones. The plant materials studied are listed in Table 1, showing the accession name i.e., the denomination by which they were included in the collection (Bartolini *et al*., 1998).

Experiment 2: Fruit samples of 'Arauco' were harvested from three sites in Mendoza Province during 2015, 2016, and 2017 growing seasons. Site 1 corresponded to the germplasm collection located in Junín and explained in Experiment 1. Site 2 corresponded to Los Campamentos, Rivadavia (33º 15'S, 68º 26' W, 660 m asl), located ~30 km southeast of Junín. The area has an average temperature of 18.5ºC, an average rainfall of 195.0 mm (mainly in summer), and a frost-free period from October to March. The olive trees were forty-years-old on average, planted at a low density (12×12 m), and surface irrigated. Site 3 corresponded to Russell, Maipú (33º 00' S; 68º 44' W, 850 m asl), located ~40 km west of Junín. The area has an average temperature of 19.5 ºC, an average rainfall of 120.5 mm (mainly in summer), and a frost-free period from October to April. The olive trees were fifty-years-old on average, planted at a low density (10×10 m), and surface irrigated.

Fruit Sample and Determination of Fruit Characteristics

In Experiments 1 and 2, three trees from each variety and site were selected, and a fruit sample of 2 kg was harvested manually around the tree canopy. All varieties were harvested when fruits started the veraison stage (changed color from yellowish-green to violet), which was visually determined. The harvest date was expressed as Julian day.

A subsample of 100 fruits was randomly selected for maturity index determination according to Beltran *et al*. (2004) by classifying fruits from 0 to 7 according to their skin and pulp color. Another subsample of 100 fruits was weighed to obtain Fruit Fresh Weight (FFW). After that, fruits were oven dried at 60ºC to constant weight to determine Fruit Dry Weight (FDW) and Moisture (M) as follows:

$$M\ (\%) = \left(\frac{FFW\ (g) - FDW(g)}{FFW\ (g)}\right) \times 100 \quad (1)$$

A subsample of 50 fruits was weighed (FFW); then, their stones were separated manually and weighed (SFW) to estimate the Pulp/Stone ratio (P/S) as follows:

$$\frac{P}{S} = \frac{FFW\ (g) - SFW(g)}{SFW\ (g)} \quad (2)$$

Fruit oil concentration was determined in three subsamples according to Avidan *et al*. (1999). In short, about 5 g of fresh pulp were weighed and placed in an oven at 60ºC until constant weight. Afterward, the dried pulp was weighed and ground in a mortar with 15 mL of petroleum ether and was transferred to a test tube. Test tubes were shaken for 12 hours and vacuum filtered with 5 mL of petroleum ether through a 100 μm filter paper. Samples were air-dried to evaporate the solvent and oven-dried at 60ºC until constant weight was obtained. The percentage of Fruit Oil Concentration (FOC) was calculated as:

$$FOC\ \% = \frac{TWO\ (g) - TWE(g)}{PW\ (g)} \times 100 \quad (3)$$

Where, TWO (g) is Tube Weight plus Oil, TWE (g) is Empty Tube Weight, and PW (g) is Pulp Weight in fresh or dry base for FOCfb and FOCdb, respectively.

Then, fruit oil content was calculated as:

Table 1. Fruit characteristics of 26 olive varieties collected over four years (2014 to 2017) from Germplasm Collection of Mendoza (Argentina). Each point is an average of three trees, except for oil concentration where nine samples were used.[a]

Variety	Harvest date (Julian day)	FOCfb (%)	CV (%)	FOCdb (%)	CV (%)	Moisture (%)	CV (%)	P/S	CV (%)	FDW (×100) (g)	CV (%)	Maturity index	CV (%)
Arauco	133	17.5 b	4.8	52.7 b	17.4	61.1 b	7.9	7.5 a	12.8	193.7 c	17.7	1.4 c	30.5
Barauni	126	19.7 a	2.8	51.7 b	6.5	61.5 b	1.0	5.7 b	12.0	256.7 b	15.3	0.9 c	22.3
Blanqueta	124	17.0 b	8.9	53.1 b	13.7	61.3 b	1.5	7.4 a	14.9	93.8 e	11.3	2.6 b	43.7
Canino	126	21.6 a	2.1	55.4 b	9.8	53.6 c	8.1	4.7 c	17.6	196.5 c	14.8	2.2 b	44.4
Chapidorado	133	14.3 b	3.6	52.4 b	14.7	61.7 b	1.6	5.6 b	6.6	151.7 d	13.2	1.0 c	6.0
Cornezuelo	130	20.0 a	6.3	56.0 b	19.8	51.4 c	3.3	3.7 c	5.1	100.5 e	5.3	2.4 b	49.5
Criolla Salvarredi	134	17.1 b	4.6	54.1 b	13.5	63.3 b	5.0	7.8 a	23.2	202.0 c	14.3	1.8 c	30.9
Criolla San Martín	131	16.4 b	6.1	52.4 b	20.1	62.8 b	5.8	6.9 a	17.7	182.4 c	12.6	1.2 c	15.2
Cucci	136	23.4 a	2.7	61.5 a	8.7	55.4 c	4.6	7.0 a	8.3	333.5 a	11.5	2.6 b	40.7
Dritta	127	18.5 a	3.4	53.1 b	8.0	55.9 c	3.1	3.7 c	7.2	90.7 e	19.5	1.7 c	52.7
Dulzal	127	18.2 a	18.2	49.0 b	17.9	55.8 c	2.4	6.2 b	28.9	168.0 d	13.3	2.9 b	49.6
Empeltre	120	14.8 b	4.6	46.7 b	13.6	59.0 b	9.4	5.7 b	13.5	189.4 c	11.4	4.6 a	20.4
Escarajuelo	118	17.7 b	1.9	51.7 b	6.1	58.4 b	3.0	6.0 b	13.6	136.3 d	22.7	1.1 c	10.8
Farga	118	18.8 a	11.5	54.1 b	12.3	50.7 c	37.8	4.7 c	9.2	119.5 e	8.3	3.3 b	13.1
Frantoio	128	22.8 a	4.7	55.7 b	19.2	49.8 c	11.5	3.8 c	10.8	121.3 e	21.6	2.7 b	55.4
Genovesa	117	19.8 a	5.0	56.8 b	14.1	59.0 b	8.4	7.1 a	19.8	239.2 b	18.0	3.1 b	24.5
Grappollo	130	19.8 a	5.0	52.0 b	12.5	53.4 c	7.5	3.5 c	5.2	81.7 e	8.1	1.5 c	44.2
Jabaluno	127	15.3 b	6.3	49.0 b	12.0	61.5 b	1.8	7.8 a	21.6	236.1 b	22.7	1.7 c	48.3
Morchiaio	116	16.0 b	47.8	44.1 b	55.3	55.3 c	11.2	3.7 c	35.7	103.4 e	19.8	3.4 b	11.9
Nebbio	119	19.9 a	3.0	54.0 b	11.0	56.3 c	9.2	5.4 b	36.0	107.2 e	13.4	1.7 c	27.9
Nevadillo Blanco	118	15.4 b	3.6	50.1 b	12.4	62.4 b	6.5	7.6 a	22.0	203.1 c	14.7	2.7 b	29.5
Nocelara	120	10.9 c	1.6	46.0 b	4.2	68.2 a	1.8	6.2 b	5.8	122.7 e	5.2	2.2 b	9.1
Piangente	117	15.7 b	4.8	48.4 b	10.4	58.1 b	7.2	4.4 c	33.6	99.5 e	18.3	3.6 b	8.2
Picual	148	21.1 a	23.7	55.5 b	23.9	55.9 c	8.8	6.7 a	17.2	216.7 c	25.1	4.2 a	20.9
Selección N°1	127	16.7 b	13.6	52.9 b	21.6	60.1 b	4.2	5.3 b	27.4	145.8 d	21.7	2.6 b	38.5
Villalonga	119	20.3 a	8.0	53.9 b	14.5	56.1 c	5.7	5.8 b	18.2	184.4 c	10.2	1.9 c	33.4
Interaction (p-value)		0.6499		0.998		0.0383		< 0.0001		0.0192		0.0192	
Year													
2014	135	20.8 a		55.2 a		54.6 d		5.9 b		184.6 b		2.9 a	
2015	119	18.6 b		52.2 b		56.4 c		7.3 a		193.6 a		3.0 a	
2016	124	16.3 c		50.9 b		59.6 b		5.8 b		144.0 d		1.6 c	
2017	115	15.6 c		51.0 b		63.9 a		4.1 c		160.0 c		2.5 b	
Interaction (p-value)		0.6499		0.998		0.0383		< 0.0001		0.0192		< 0.0001	

[a] FOCfb: Fruit Oil Concentration in fresh base; FOCdb: Fruit Oil Concentration in dry base; P/S: Pulp/Stone ratio; FDW: Fruit Dry Weight; CV: Coefficient of Variation. (a-c) Values with the same letter are not significantly different among years by DGC test at P≤ 0.05. Significant differences are indicated with different letters.

$$Fruit\ oil\ content = FOCdb \times$$
$$(FDW - (FDW/(P/S))/100) \qquad (4)$$

Calibration and Validation of a Simple Method for Fruit Oil Content Estimation

Regression analysis was applied between fruit oil content and FPDW using the data compiled during 2014, 2015, and 2016 from all the assays described above (n= 242). To validate the model, only independent data from 2017 of ten varieties from the olive germplasm collection of Junín and data of 'Arauco' from Rivadavia and Maipú were used (n= 42).

The correspondence between observed and estimated fruit oil content values was graphically compared with the expected 1:1 equation and Root Mean Square Error (RMSE) using Irene software beta version. The RSME represents the mean distance between prediction and measurement.

Statistical Analysis

ANOVA was used to test the effect of variety, year, and sites on the response variables. Means were separated using the DGC-test (Di Rienzo *et al*., 2002) for a level of significance of α= 0.05. Statistical analysis was performed using the InfoStat 1.5 program and regression analysis was done using the GraphPad Prism version 5.01 software.

Significant difference (P≤ 0.05) on measured variables for variety, year, and sites were determined by ANOVA and separated using the DGC-test (Di Rienzo *et al*., 2002). Statistical analysis was performed using the InfoStat 1.5 program and regression analysis was done using the GraphPad Prism version 5.01 software.

RESULTS

Fruit Characteristics

The fruit characteristics evaluated in 26 varieties from the olive germplasm collection of Mendoza during four seasons

are shown in Table 1. Varieties (P≤ 0.01) and years (P≤ *0. 01*) had a significant influence on all evaluated characteristics of fruit. While variety×year interaction was not significant for FOCfb and FOCdb, it was significant for the rest of the fruit characteristics (Table 1).

Across all varieties, fruit oil concentration on both fresh and dry base was highest during 2014 (Average FOCfb= 20.8% and FOCdb= 55.2%) and lowest during 2017 and 2016 (average FOCfb= 15.6 and 16.3 % and average FOCdb= 51.0 and 50.9%, respectively). FOCdb was not significantly different from 2015 to 2017. In 2014, fruits showed the least moisture (54.6%), while the highest (63.9%) was observed in the 2017 harvest. The highest Maturity Index (MI) was recorded in 2014 and 2015 (2.9 and 3.0, respectively), while the lowest value (1.6) occurred in 2016. On average for all varieties, fruits were heaviest (193.6 g) during 2015, intermediate (184.6–160.0 g) in 2014, and 2017 respectively, and lightest (144.0 g) in 2016. The P/S was highest (7.3) in 2015, intermediate (5.8 and 5.9) in 2016 and 2014 respectively, and lowest (4.1) in 2017. On average across the four seasons, varieties were harvested from 116 to 148 Julian days. The harvest period showed a maximum variation of twenty days between the earlier harvest year (2017) and the later harvest year (2014).

The varieties evaluated showed a smaller FOCdb variation range compared with the rest of the fruit characteristics. Varieties were statistically separated in two groups for FOCdb; in three groups for FOCfb, moisture, pulp/stone ratio, and maturity index; and in five groups for fruit dry weight.

Fruit oil concentration in fresh base varied significantly among varieties from 10.9% to 23.4 % with an average of 17.8 % for the 26 varieties. Based on FOCfb, varieties were grouped in three clusters: 1) Varieties with FOCfb > 18%: 'Barauni', 'Canino', 'Cornezuelo', 'Cucci', 'Dritta', 'Dulzal', 'Farga', 'Frantoio', 'Genovesa', 'Grappollo', 'Nebbio', 'Picual', and

'Villalonga' from now on "Highest FOCfb"; 2) Varieties with medium fruit oil concentration (18 % > FOCfb > 14%: 'Arauco', 'Blanqueta', 'Chapidorado', 'Criolla S', 'Criolla SM', 'Empeltre', 'Escarajuelo', ''Jabaluno', 'Morchiaio', 'Nevadillo Blanco', 'Piangente', and 'Selección Nº1'); and 3) Only 'Nocelara', with the lowest oil concentration in fresh base (FOCfb < 11%).

Fruit oil concentration in dry base ranged from 44.1 to 61.5%, where 25 out of 26 varieties belonged to the same group (< 56%). 'Cucci' had FOCdb= 61.5%, significantly higher than the rest of the varieties. Even comparing among years, except for 2014, FOCdb was not significantly different among harvests from 2015-2017.

Fruit moisture presented a wide range of variation from 49.8% to 68.2%. Except for 'Morchiaio', all varieties with the lowest moisture (< 57%) were also present in the "Highest FOCfb" cluster. The varieties with the highest FOCfb (> 18%) and lowest moisture (< 57%) were 'Canino', 'Cornezuelo', 'Cucci', 'Dritta', 'Dulzal', 'Farga', 'Frantoio', 'Grappollo', 'Nebbio', 'Picual', and 'Villalonga'.

The P/S ratio ranged from 7.8 to 3.5. Based on the P/S ratio, varieties were clustered into three groups: varieties with the highest ratio (P/S> 6.5), intermediate ratio (6.5> P/S> 5), and lowest ratio (P/S< 5). 'Cucci' and 'Picual' were highlighted for their high P/S ratio from the "Highest FOCfb" group with no difference from 'Arauco', 'Blanqueta', 'Criolla S', 'Criolla SM', 'Genovesa', 'Jabaluno' and 'Nevadillo Blanco'.

Fruit dry weight over 100 fruits showed a wide range between 333.5 g and 81.7 g. Based on FDW, varieties were clustered in five groups: "super-high" > 300 g, "high" from 300 to 230 g, "elevated" from 230 to 180 g, "medium" from 180 to 130 g, and "low" < 130 g, respectively. 'Cucci' was the only variety present in the "super-high" cluster (333.5 g) and was highlighted from

the "Highest FOCfb" cluster. 'Grappollo' was in the lowest cluster (81.7 g), but it was not significantly different from 'Blanqueta', 'Cornezuelo', 'Dritta', 'Farga', 'Frantoio', 'Morchiaio ', 'Nebbio', 'Nocelara', and 'Piangente'.

The maturity index ranged widely from 0.9 to 4.6. Based on MI, varieties were clustered into three groups: MI< 2, MI from 2 to 4, and MI> 4. Only 'Picual' showed a high MI from the varieties in the "highest FOCfb" cluster, while 'Barauni', 'Dritta', 'Grappollo', 'Nebbio', and 'Villalonga' had the lowest MI.

Environmental Influence on 'Arauco' Variety Fruit Characteristics

On average, for years 2015, 2016, and 2017, the fruit characteristics measured in the 'Arauco' did not show any significant differences among Junín, Rivadavia, and Maipú study sites. Moreover, 'Arauco' was harvested from three sites and for three years at 133 J on average. In contrast, growing seasons markedly affected FOCfb, M, P/S ratio, FDW and MI (Table 2). Fruits collected during 2015 presented the highest FOCdb (51.0%), FDW (196.1 g 100 fruits^{-1}), MI (2.1), P/S ratio (7.5), and the lowest M (61.0%). Contrary to this, fruits harvested in 2017 showed the lowest FOCfb (14.0%), FOCdb (48.1%), MI (1.5) and P/S ratio (4.9).

Calibration and Validation of a Model to Estimate Fruit Oil Content

Pooling data of all varieties from years 2014, 2015, and 2016, a simple method to estimate fruit oil content was calculated (Figure 1). The model was adjusted using a large number of samples (n= 242) and a wide range of pulp dry weights (0.5-3.5 g). A single and positive relationship described the association between fruit oil content (g

oil/fruit) and fruit pulp dry weight (FPDW). Fruit oil content was estimated at -0.05+0.56xFPDW; r= 0.99. The model showed a RSME= 0.06. Consequently, the fruit oil content was estimated from FPDW data with high efficiency (0.95).

The validation explained almost 96% of the data, a low RSME= 0.05 and a high correlation coefficient (r= 0.98) were observed (Figure2).

DISCUSSION

Fruit Characteristics

Fruit characteristics enabled clustering varieties with agronomic or industrial benefits. FOCfb is used worldwide by oil makers to determine the oil yield (Beltrán *et al*., 2004; Zipori *et al*., 2016). In Argentina, instead, most industries pay farmers a fixed percentage of oil yield, and farmers charge that percentage in relation to kilogram of fruit. Trentacoste *et al*. (2016), evaluating 'Arbequina', determined that FFW depended on water and oil concentration. A decrease in fruit moisture was proportional to an increase in oil concentration. As shown in Table 1, most varieties with high FOCfb had low moisture. The same trend was observed for annual averages; for instance, 2014 presented the highest FOCfb and the lowest moisture, while 2017 presented the lowest FOCfb and the highest moisture. These results are important from an industrial viewpoint because varieties with high moisture during malaxation produce emulsions (De la Rosa *et al*., 2008) reducing oil extractability (Motilva *et al*., 2000). In our results, eleven varieties showed high FOCfb and low moisture ('Canino', 'Cornezuelo', 'Cucci', 'Dritta', 'Dulzal', 'Farga', 'Frantoio', 'Grappollo', 'Nebbio', 'Picual', and 'Villalonga'), from now on referred to as 'oil varieties'. Eight of them are not commonly grown in Argentina.

Table 2. Fruit characteristics of 'Arauco' variety collected from three sites over three years (2015 to 2017) in Mendoza Province (Argentina). Each point is an average of three trees, except for oil concentration where nine samples were used.[a]

Site	Harvest date (Julian day)	FOCfb (%)	CV (%)	FOCdb (%)	CV (%)	Moisture (%)	CV (%)	P/S	CV (%)	FDW (×100) (g)	CV (%)	Maturity index	CV (%)
Junín	130	15.8 a	16.3	51.6 a	4.1	63.8 a	4.3	7.9 a	11.3	182.3 a	19.2	1.2 a	30.9
Maipú	138	15.7 a	54.2	50.4 a	7.9	63.0 a	2.8	7.7 a	22.9	184.4 a	27.7	2.3 a	73.7
Rivadavia	131	15.1 a	16.5	48.2 a	9.6	63.0 a	4.3	7.5 a	13.4	189.8 a	14.9	1.8 a	42.4
Year													
2015	134	17.5 a	12.4	51.0 a	10.4	61.0 b	3.4	7.5 b	19.1	196.1 a	18.6	2.1 a	61.8
2016	138	15.2 a	13.9	48.9 a	9.1	64.2 a	2.7	7.5 b	10.0	178.7 b	17.9	1.8 b	49.6
2017	129	14.0 a	45.3	48.1 b	4.8	64.2 a	3.8	7.8 a	16.5	186.9 b	18.5	1.5 b	47.7
Interaction (P-value)		0.6555		0.0927		0.0002		< 0.0001		< 0.0001		< 0.0001	

[a] FOCfb: Fruit Oil Concentration in fresh base; FOCdb: Fruit Oil Concentration in dry base; P/S: Pulp/Stone ratio; FDW: Fruit Dry Weight; CV: Coefficient of Variation. (a-b) Values with the same letter are not significantly different among years by DGC test at P≤ 0.05. Significant differences are indicated with different letters.
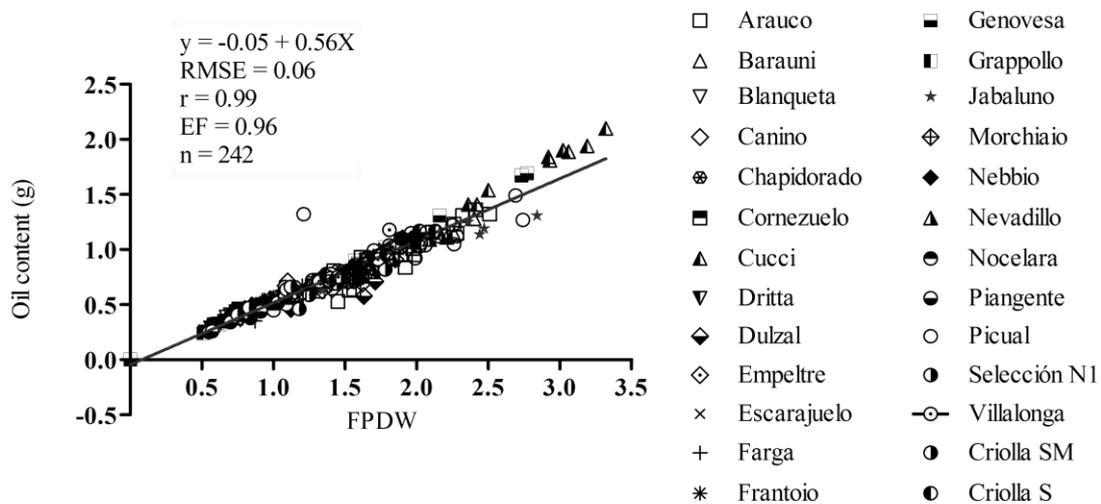
Figure 1. Model calibration. Relationship between fruit oil content and Fruit Pulp Dry Weight (FPDW) from 26 varieties and 3 sites over 3 years**.** RMSE is the Root Mean Squared Error. And EF is the Efficiency of the model.
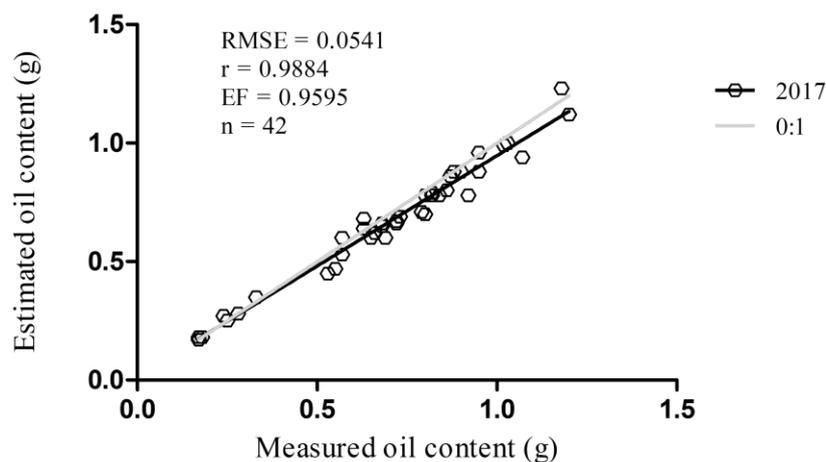

Figure 2. Model validation. Relationship between measured and estimated fruit oil content in 2017 of ten varieties from the collection and of 'Arauco' from 3 environments. RMSE is the Root Mean Squared Error. EF is the efficiency of the model validation.

The slight FOCdb variability among the varieties and environmental conditions studied could be explained because the initial population was selected according to high FOCdb and low moisture from a larger population (74 accessions) by Trentacoste and Puertas (2011). For that reason, negligible differences for FOCdb were observed among the varieties studied, except for 'Cucci', which was also already highlighted by the same authors. Thus, FOCfb variations were explained by moisture rather than by fruit oil accumulation, which allowed developing a robust model to estimate oil content through pulp dry weight.

Taking into account that more than 90% of oil is in the pulp (Beltrán *et al*., 2003), fruit oil concentration could be expressed based on pulp weight, ignoring the little oil content

in fruit stone. There is also a close association between fruit dry weight and fruit oil content (Rondanini *et al.*, 2014). Determining P/S ratio and FDW could provide important data for the olive industry because it would allow evaluating and clustering many varieties in a short time by their high oil content. In addition, varieties with high P/S ratio and FDW could be used for table olives or for a double purpose (Giuffrè, 2017). In this study, the average P/S and FDW for all varieties and years were 5.7 and 164.5 g/100 fruits, respectively. The 'Criolla S' showed the highest P/S= 7.8, whereas the 'Cucci' showed the highest FDW= 333.5 g 100 fruits^{-1}. In contrast, 'Grappollo' showed the lowest values (P/S= 3.5 and FDW= 81.7 g 100 fruits^{-1}). FDW is also taken into account for mechanical harvesting efficiency because shakers remove the heaviest fruit better (Beltrán *et al.*, 2004). In addition, FDW, as well as P/S ratio, are taken into account in breeding programs as significant olive characteristics (Tous *et al.*, 2011).

In Junín, the frost-free period spans from November to April. Even though the olive tree is cold-tolerant, frosts trigger damage in fruits, which leads to low quality oils (Morelló *et al.*, 2003). Fruit color (MI≥ 3) is a widely used index to determine harvest day (Beltrán *et al.*, 2004). However, some varieties never reach that value before the occurrence of a frost event. Taking this into account, early and late varieties were classified using maturity index and harvest date (J). In our results, there were non-significant differences between the MI of 'Empeltre' (MI= 4.6, 120 J) and 'Picual' (MI= 4.2, 148 J). Nevertheless, 'Empeltre' can be considered as an early variety because it is harvested twenty-eight days before 'Picual'. Similarly, varieties with low maturity index such as 'Dritta' (MI=1.7, 127 J), 'Grappollo' (MI= 1.5, 130 J), 'Nebbio' (MI= 1.7, 119 J), and 'Villalonga' (MI= 1.9, 119 J) can be considered late varieties. Except for 'Empeltre', all these varieties were selected from the "oil varieties" cluster.

As shown in Table 1, a significant difference was observed in the MI for all growing seasons. The lowest and highest MI were observed in 2016 and 2015, respectively, and the average of four years was MI= 2.5 (data not shown). Differences among years may be due to variation in crop load, as previously demonstrated by Trentacoste *et al.* (2010). Crop load was not measured in this work, however, the olive tree cycle consists of a low-yield crop year followed by a high-yield crop year known as "alternate bearing" (Lavee, 2007).

Environmental Influence on 'Arauco' Fruit Characteristics

In Argentina, 'Arauco' is the only autochthonous variety. It has been widely destined to table olive elaboration due to its low oil content, sizeable fruit fresh weight (one of the biggest among regionally-cultivated varieties), and high P/S ratio, higher than the average of all the introduced varieties evaluated here. 'Arauco' showed an average P/S ratio across sites of 6.7, higher than 5.7 for the other varieties. Similarly, fruit dry weight from 'Arauco' versus 'the other varieties' was 185.5 and 164.5 g 100 fruits^{-1}, respectively. In addition, 'Arauco' presented a lower fruit oil concentration than the average of the other varieties studied here. Thus, on average, FOCfb and FOCdb for 'Arauco' versus 'the other varieties' were 15.5 vs 18.0% and 50.1 vs 52.4%, respectively. 'Arauco' presented, on average, a low MI compared with the rest (1.8 and 2.5, respectively), which, along with harvest date, proved the late maturity of 'Arauco'.

Calibration and Validation of a Model to Estimate Fruit Oil Content

Olives should be harvested when fruit oil content has reached its maximum. According to Mickelbart and James (2003), maximum oil accumulation is not related to fruit color. It

would be possible to harvest fruits at maximum oil content by measuring oil accumulation rather than fruit color during fruit ripening. According to Beltrán (2003), fruit oil content shows high genetic variability, especially in dry base. This could allow classifying cluster varieties by their potential oil extraction yield.

Pooling the data from all varieties, environments, and growing seasons in a single relationship described the high association between fruit oil content and fruit dry weight ($R^2 = 0.99$). Given that in this method pulp dry weight is fast and easy to estimate, many samples could be analyzed in a short time. For our determinations, only an oven and a scale were enough, while other research works were performed by means of NIRS (near-infrared reflectance spectroscopy), a costly laboratory equipment. In addition, analyzing several samples in a short period of time is useful for comparative research assays (Trentacoste *et al.*, 2018).

In model calibration, the data presented a wide range of oil content in dry base (44.0−61.5%) and fruit dry weight (81.7-333.3 g 100 fruits^{-1}), which allowed developing a widely consistent calibration model. Validation of the model showed its robustness ($R^2 = 0.99$) allowing the estimation of reliable data.

In the industry, oil yield is calculated as an established percentage of fresh olive weight, and it is reimbursed to the farmers. According to Zipori (2016), oil content in fresh base seems to be unreliable, given that it often fluctuates due to environmental factors. This model allows estimating reliable oil content and possibly predicting oil yield with little error. Another application of the model might be to determine optimal harvest time, measuring fruit during ripening when it has reached its maximum oil content.

CONCLUSIONS

Eleven varieties were highlighted from the collection for their high FOCfb and low moisture. Eight of them, scarcely grown in Argentina, expressed good fruit characteristics allowing us to assume that they were well adapted. Characterizing varieties in local environments allows increasing the genetic variability available or discovering varieties better adapted than those traditionally grown.

The 'Arauco' fruit characteristics were very stable among the environments studied, which could be considered an adaptive response to local conditions. Among the varieties evaluated, 'Arauco' shows a medium fruit oil concentration in both fresh and dry base, and a high weight of dry fruit, which turns it into an excellent double-purpose variety. The characterization of autochthonous varieties has been used as an important tool for developing Protected Designations of Origin.

The model would allow calculating the best harvest time simply by determining fruit dry weight. We propose fruit dry weight as a reliable estimator of olive oil content in fruit, given the simplicity and practicality of this technique, which would allow processing hundreds of samples per day in a laboratory. In addition, it would provide a better fruit oil estimator for farmers and producers. Lastly, this technique represents an economic and fast method when it comes to estimating fruit oil content in a large number of samples, as in the case of olive breeding programs.

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ویژگی های میوه و برآوردی ساده از محتوای روغن در بیست وشش ژنوتیپ زیتون در آرژانتین

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چکیده

در سطح جهان، بیش از دوهزار رقم زیتون وجود دارد که بیشتر آنها بومی منطقه مدیترانه است. با اینهمه، فقط تعداد محدودی از ارقام زیتون در آرژانتین کشت می شود. در استان مندوزا، کلکسیونی وجود دارد که شامل بیش از ۷۰ نمونه ثبت شده می باشد، با این وجود، اطلاعات در مورد سازگاری واریته های مختلف زیتون به اقلیم خشک این استان کم است. اهداف این پژوهش شامل ارزیابی اهمیت زراعی ویژگی های میوه در ۲۶ رقم و ژنوتیپ این کلکسیون و مقایسه ویژگی های یک رقم محلی به نام Arauco در شرایط مختلف محیطی و فصل رشدهای متفاوت در مندوزا بود. افزون بر این، با استفاده از طیف متنوعی از رقم های زیتون، یک روش اقتصادی ساده و سریع برای تعیین محتوای روغن میوه کالیبره شد و راستی آزمایی گردید. نتایج آزمایش در باره همه ویژگی های ارزیابی شده تفاوت های معنادار(*0.01* ≥P) بین رقم ها و سالهای رشد نشان داد. محتوای روغن میوه رابطه نزدیک ومثبتی با وزن خشک تفاله میوه داشت (0.99 =r ;0.56x+ 0.05- =y). از نظر بالا بودن غلظت روغن بر مبنای وزن تر و کم بودن مقدار رطوبت، ارقام برتر کلکسیون شامل بودند بر: ,'Canino' 'Cornezuelo', 'Cucci', 'Dritta', 'Dulzal', 'Farga', 'Frantoio', 'Grappollo',

‘Nebbio’, ‘Picual’, and ‘Villalonga’. افزون بر این، همه ویژگی های ارزیابی شده میوه Arauco در محیط های مورد مطالعه مشابه بود و به طور معناداری تحت تاثیر شرایط فصل رشد قرار داشت. مدل پیشنهادی که با داده های مستقل کالیبره و راستی آزمایی شد به سادگی و با تجزیه چندین نمونه در زمانی کوتاه با استفاده از وزن خشک تفاله ، بهترین زمان برداشت و مقدار محتوای روغن را تعیین می کند.